



in situ Rolling Circle Amplification (*in situ* RCA) for Genetic Diagnostics

Abstract

The invention is an improved *in situ* rolling circle amplification (RCA) for DNA or RNA amplification in isothermal conditions. The efficiency of the improved *in situ* RCA is about 90%. It can be used to detect *in situ* tissue DNA sequence, gene expression or single nucleotide polymorphisms (SNP). Applications of *in situ* RCA include cancer diagnosis, population identification for risk assessment, disease susceptibility, or medical treatments, and other medical and veterinary diagnostics. By using this *in situ* RCA technology, scientists at LLNL have successfully identified cancer cells in tissues, detected DNA and RNA mutations and quantified RNA expression levels on cells and tissues directly.

This *in situ* RCA is simple, easy, and sensitive. A few steps are added to the normal *in situ* RCA to improve the efficiency. It can be carried out with conventional clinic laboratory conditions and equipments with reagents and equipments commercially available. One copy of either DNA or RNA is sufficient for the detection. Because the resulting amplification product is large and anchored to the target, the product remains inside the cell during the process and the amplification product does not diffuse to surrounding cells or solutions, cells or tissues containing the specific DNA or RNA can be readily identified. Alternatively, the process of this *in situ* RCA can be automated for high throughput tissue examinations.

Patents and Publications:

US Patent 6,783,943, Rolling circle amplification detection of RNA and DNA, Christian; Allen T., et al.

Christian, A. T. ; Pattee, M. S.; Attix, C. M.; et al. Detection of DNA point mutations and mRNA expression levels by rolling circle amplification in individual cells, Proc. Natl. Acad. Sci. USA, Vol. 98, Issue 25, 14238-14243, December 4, 2001.
<http://www.pnas.org/cgi/content/full/98/25/14238>.

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